IOSR Journal of Dental and Medical Sciences (IOSR-JDMS)
e-ISSN: 2279-0853, p-ISSN: 2279-0861.Volume 15, Issue 4 Ver. I (Apr. 2016), PP 100-104
www.iosrjournals.org

Hba1c as a Diagnostic Criteria in Diabetes Mellitus A Comparison with Mini-Ogtt

Dr Swati Tiwari, Dr Vaishali Jain, Dr S.S.Biswas, Prerna Vyas Gokhale
(Department of Biochemistry, L.N. Medical College, Kolar Road, Bhopal, MP, India)

Abstract: *HbA_1c is not merely a marker for monitoring diabetes but has been accepted as a marker for its diagnosis too. The diagnostic efficiency of the 6.5 % cutoff recommended by ADA was evaluated against the standard mini oral glucose tolerance test (OGTT) in our local population to find an optimum cutoff of HbA_1c in Bhopal. Estimation of HbA1c, fasting plasma glucose and 2 hour post 75 gms oral glucose challenge plasma glucose was done in 270 subjects. HbA_1c diagnosed comparable but insignificantly fewer number of patients as diabetic ($p>0.05$) but significantly more as pre-diabetic.($p<0.001$). ROC analysis of various HbA_1c cutoffs against the standard mini-OGTT showed 60% sensitivity and 92% specificity at recommended cutoff of $HbA_1c>= 6.5$. Negative Predictive Value (NPV) and Positive Predictive Value (PPV) were found to be 83% and 77% respectively. The optimum sensitivity of 72% and specificity of 82% was obtained at HbA_1 cut off 6.1; for which NPV and PPV were found to be 85% and 57% respectively. In conclusion, HbA_1c proved to be slightly inferior but comparable to mini-OGTT in detecting diabetes. The use of the lower optimum cutoff 6.1 is recommended in this population to increase the yield of diabetics and minimize the gap with mini-OGTT. HbA_1c detected significantly more pre-diabetics than mini-OGTT, hence it may be used to screen the local population.*

Key words: *Diabetic, Prediabetic, Glycosylated haemoglobin, Oral Glucose Tolerance Test.*

I. Introduction

Asia has been the epicenter of diabetes in the world, with India and China leading the way in the prevalence of diabetes. In 2000, India had more people (31.7 million) with estimated diabetes mellitus than any other country in the world, followed closely by China (20.8 million). (1) China has since overtaken India as the diabetic capital of the world. It has about 92 million diabetics which is far more than the 62 million estimated in India. (2) However, in some urban areas of south India, prevalence of diabetes is as high as nearly 20%. (3).

The gold standard in the diagnosis of diabetes has been the oral glucose tolerance test (OGTT). However, the oral load of glucose and the half hourly sample collection makes it a difficult test to carry out in such large numbers. Using simply the fasting plasma glucose (FPG) for the diagnosis of diabetes has its disadvantages, as it often misses out on cases of diabetes detected by the post challenge plasma glucose (PCPG), besides fasting itself is an inconvenience. (4) Glycosylated hemoglobin (HbA_1c) does not require fasting, and hyperglycemia being the cause of complications of diabetes, a long term measure of hyperglycemia (HbA_1c) is a better marker of the severity of the disease than a single measurement of glucose. (5)

An International Expert Committee (IEC) with members appointed by the American Diabetes Association (ADA), the European Association for the Study of Diabetes, and the International Diabetes Federation recommended that HbA_1c > 6.5 % be considered as diagnostic of diabetes. (5) Their recommendation was based on the evidence of ROC (Receiver Operating Characteristic) analysis data that the cut point of HbA_1c for diabetes specific retinopathy was 6.5%. Retinopathy has widely been accepted as the best criterion for comparing glycaemic measures because it is a specific, objective, and relatively early clinical complication of diabetes. (6)

In 2010, the ADA accepted this criterion. They recommended that HbA_1c > 6.5% be included as a criterion for diagnosis of diabetes, and people with HbA_1c between 5.7-6.4% be considered at risk of developing diabetes in the future. (7) In 2011, the WHO gave a conditional acceptance to the use of HbA_1c for the diagnosis of diabetes subjecting it to stringent quality assurance tests and standardization criteria, besides mandating the exclusion of conditions which precludes its accurate measurement. (8)

HbA_1c is known to be affected by ethnicity, irrespective of the state of glycaemia. (9) (10) Blacks have widely been reported to have higher levels of HbA_1c than whites. (11) Asians and American Indians have also been reported to have higher levels of HbA_1c than local whites. (12)

This study was therefore carried out in our local population in Bhopal to find out the diagnostic efficiency of the recommended cutoff of $HbA_1c>= 6.5$ % against the standard mini OGTT and to find an optimum cutoff of HbA_1c in our population.

II. Material And Methods

This study was a hospital based cross-sectional study conducted on 270 men and women visiting IPD/OPD of Medicine department of the J.K. Hospital and LN Medical College, Kolar, Bhopal. The study group included 162 males and 108 females who were between 18 to 80 years of age. They included 59 Young (<40 years), 107 Middle aged (40-59 years) and 104 Old people (>= 60 years). A structured questionnaire was used to seek general details of the subject's economic status, diet, smoking, alcohol consumption, medical and drug history. Those excluded from the study were already diagnosed cases of diabetes, pregnant women, patients taking medication causing rapid rise in plasma glucose, patients of hemolytic anaemia, iron deficiency anaemia and hemoglobinopathies, patients with hemoglobin variants, history of chronic blood loss or recent blood transfusion. Study was conducted after clearance by Institutional Ethics Committee and after obtaining informed consent of the subjects. Sample size was calculated on the basis of a previous study. (13)

Modified mini OGTT was performed taking only two venous blood samples, one after overnight fasting (10-12 hours) and the other two hours after a 75 Grams of oral anhydrous glucose load (in 300 ml water). The fasting samples were used for the estimation of FPG and HbA1c. Fluoride vials were used to collect samples for glucose estimation and EDTA vials for HbA1c. All parameters were analyzed on fully automated analyzer. Glycosylated hemoglobin was estimated using turbidimetric inhibition immunoassay and glucose was estimated using glucose oxidase/ peroxidase method.

The data obtained was analyzed using software IBM SPSS version 16, Chicago, USA. The statistical tests used were chi-square test, odds ratio and Pearson's correlation. The difference between groups for the above tests was considered significant if the P value was less than 0.05. ROC (Receiver Operating Characteristic) analysis was done to assess sensitivity and specificity of the recommended cutoff of HbA_1c against the standard mini-OGTT and also to decide on an optimum cutoff value of HbA_1c in our population for the diagnosis of diabetes. Negative and positive predictive values (NPV and PPV) and likelihood ratios (LR +ve and LR-ve) were calculated for each cutoff.

III. Results

Subjects diagnosed diabetic by mini-OGTT were slightly more than by HbA_1c, but it was not statistically significant (p=0.122). In contrast, the number of patients diagnosed pre-diabetic were significantly more by HbA_1c (p<0.001), while the number found to be normal were significantly more by mini-OGTT (p<0.001). [Table 1] HbA_1c showed significant association with mini-OGTT in diagnosing diabetics ($\chi 2 = 112$, p value <0.001)

The odds of diagnosing diabetic to not diagnosing diabetic were 44% more when diabetes was diagnosed by mini-OGTT than by HbA_1c. However, this difference was not statistically significant (p = 0.08). The odds of diagnosing pre-diabetes to normal were 90% less by mini-OGTT than by HbA_1c. This difference was statistically significant (p<0.001). [Table 2]

Significant (p<0.001) correlation of HbA_1c was found with both FPG (r=0.744) and PCPG (r=0.766). [Table 3] Scatter plot of FPG with HbA_1c [Fig 1] was better than that of PCPG with HbA_1c [Fig 2] although both confirmed the linear relationship.

ROC curve with mini-OGTT as the standard test for diagnosing diabetes and HbA_1c as the screening test showed Area Under the Curve 0.822 and p value <0.001, showing that HbA_1c was a significant predictor of diabetes as diagnosed by OGTT. [Fig 3] ROC analysis of various HbA_1c cutoffs against the standard mini-OGTT showed 60% sensitivity and 92% specificity at the ADA recommended cutoff of 6.5. LR+ was 3.4 and LR- was 0.20 at that level. The optimum specificity and sensitivity of 72% and 82% respectively was obtained at HbA_1c cutoff 6.1. [Table 4]

Table 1 Comparison of diabetic status by mini-OGTT and HbA1c

Diabetic status	Mini-OGTT	HbA1c	$\chi 2$	p-value
Normal	159	95	16	<0.001
Pre-diabetic	26	109	51	<0.001
Diabetic	85	66	2.4	0.122

Table 1 Odds Ratio for chance of diagnosing diabetic and pre-diabetic by two criteria

Category	Odds by Mini-OGTT	Odds by HbA1c	Odds Ratio	95% CI	p-value
Diabetic/Non-diabetic	0.46	0.32	1.44	0.61-2.11	0.08
Pre-diabetic/Normal	0.16	1.65	0.10	0.05-0.15	<0.001

Table 3 Correlation of HbA1c with FPG and PCPG

Test parameter1	Test Parameter 2	r	P value
HbA1c	FPG	0.744	<0.001
HbA1c	PCPG	0.766	<0.001

Table 4 Performance of different HbA1c cutoffs against mini-OGTT

HbA1c Cutoff (gm%)	Sensitivity	Specificity	Accuracy	NPV	PPV	LR+	LR-
5.7	82%	54%	58%	88%	42%	0.73	0.13
5.8	80%	62%	62%	86%	44%	0.81	0.15
5.9	74%	67%	67%	87%	49%	0.97	0.15
6.0	71%	76%	69%	85%	50%	1.03	0.17
6.1	**72%**	**82%**	**74%**	**85%**	**57%**	**1.35**	**0.17**
6.2	65%	87%	78%	86%	64%	1.79	0.16
6.3	63%	90%	79%	84%	68%	2.2	0.18
6.4	60%	92%	81%	83%	74%	2.9	0.19
6.5 (ADA)	**60%**	**92%**	**82%**	**83%**	**77%**	**3.4**	**0.20**
6.6	56%	95%	82%	83%	78%	3.6	0.19
6.7	54%	96%	82%	82%	84%	5.3	0.21

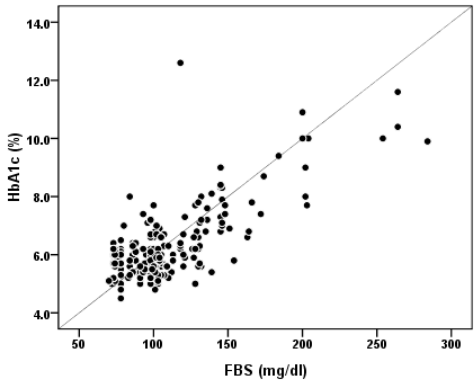

Figure 1: Scatter plot of HbA1c with FPG

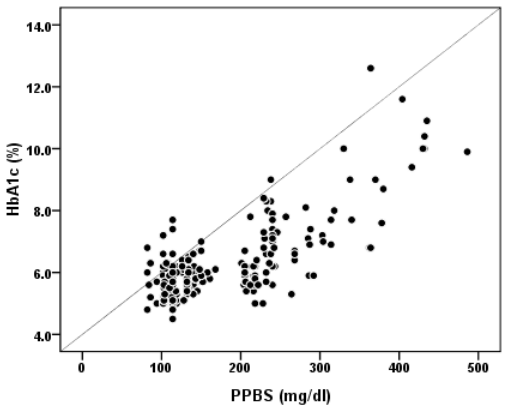

Figure 2: Scatter plot of HbA1c with PPBS

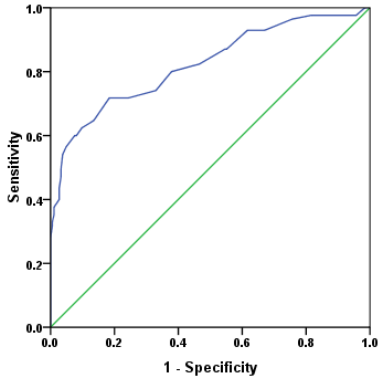

Figure 3: ROC curve with mini-OGTT as standard test for diagnosing diabetes and HbA1c as screening test

IV. Discussion

HbA_1c fared well against mini-OGTT in diagnosing diabetes as it diagnosed an almost similar number i.e. 66 as diabetic (24%) to 85 (31%) by mini-OGTT. Of these, 55 were diagnosed diabetic by either criterion. The odds of diagnosing diabetic/non-diabetic were 44% higher by mini-OGTT, but again not statistically significant. Hjellestad et al reported in Norwegian patients that HbA_1c diagnosed a similar percentage (14.6%) as diabetic as OGTT (12%). (14) Incani et al reported in Italian participants that prevalence of diabetes by the two criteria was similar i.e. 28% by OGTT and 24% by HbA_1c in an obese cohort and 11% by both OGTT and HbA_1c in a diabetes screening cohort. (15) This trend is largely followed for HbA_1c cutoff 6.5% but gets reversed on taking the HbA_1c cutoff higher. Raman et al took HbA_1c cutoff 7% as a treatment limit in an undiagnosed group at high risk of developing diabetes and reported significantly more diabetics (49%) by HbA_1c compared to OGTT (17%). (16)

HbA_1c proved comparable to mini-OGTT by other measures too. It showed significant association with mini-OGTT in diagnosing diabetics and was a significant predictor of diabetic status by mini-OGTT (AUC was 0.82) in ROC analysis. Hjellestad et al reported similar but slightly lower AUC (0.73) for ROC analysis of HbA_1c against OGTT. (14) In this study, HbA_1c had high LR+ i.e. 3.4 and low LR- i.e. 0.20, so it showed high probability of being positive and low probability of being negative in diabetics. HbA_1c expectedly showed significant correlation with fasting and post-challenge 2 hour plasma glucose (r=0.744 for fasting and r=0.766 for post challenge plasma glucose). Studies by Riet et al and Ghazanfari et al reported similar correlation of HbA_1c with FPG and PCPG, the correlation being stronger in cases of known diabetes than otherwise. (17) (18)
HbA_1c diagnosed 109 (40%) prediabetics as compared to 26 (10%) by mini-OGTT, which was significantly higher (p-value<0.001). Hjellestad et al also reported twice the amount of pre-diabetic by HbA_1c than by OGTT (70% vs 33%) in his study on Italian patients. (14) Rivers et al also reported in a study on Bahamian adolescent students a much higher yield of pre-diabetics by HbA_1c (16%) compared to OGTT (1%), attributing fluctuations to hormonal changes. (19) Incani et al, reported conversely a higher yield of prediabetics by OGTT rather than by HbA_1c i.e. 65% to 43% in the diabetic screening cohort and a similar yield i.e. 29% to 27% respectively in the obese cohort. (15)

In this study, the ADA recommended cutoff of 6.5 HbA_1c showed very good specificity (92%) but relatively modest sensitivity (60%). NPV was slightly higher than PPV i.e. 83% and 77% respectively. In a large cross sectional study among adults in Chandigarh, Kumar et al reported similar results with HbA_1c >= 6.5% against OGTT having specificity 88%, sensitivity 65%, NPV 96% and PPV 75%. (20) Studies by Hjellestad et al and Riet et al have confirmed to this trend of lower sensitivity and higher specificity at HbA_1c >= 6.5%. (17) Incani et al reported results similar to our study in an obesity clinic cohort, but lower sensitivity and PPV in a diabetes screening cohort. (15) Engelgau et al used diabetic retinopathy to define diabetes instead of OGTT in a cross section of Egyptians. At HbA_1c >= 6.5%, they reported sensitivity 51% and specificity 86% in a subpopulation not receiving antihyperglycaemic medication, while sensitivity was 79% and specificity was 75% in the total population. (21) Their findings in the untreated subpopulation are close to the findings in this study. Overall, most studies reported this trend of sensitivity lower than specificity and PPV lower than NPV for HbA_1c >= 6.5%, although the figures varied from one study to another depending on the population studied and the methodology.

In this study, the optimum sensitivity and specificity, i.e. 72% & 82% were obtained at HbA_1c >= 6.1%, which is below the ADA and WHO recommended cutoff of 6.5%. Reports on the optimum cutoff for HbA_1c for diagnosing diabetes have varied worldwide, 5.5% in the Gomyo study (22), 5.8% in the Dutch general population, (17) 6.0% in The Diabetes Control and Complications Trial (DCCT), (23) 6.2% in The United Kingdom Prospective Diabetes Study (24). The Australian Diabetes Society Expert Committee agreed with the recommended HbA_1c cutoff >= 6.5%, but recommended that lower than 6.5% HbA_1c did not rule out a diagnosis of diabetes. The New Zealand Society for the Study of Diabetes favoured the use of HbA_1c at >= 6.7% for the diagnosis of diabetes ahead of OGTT in 2011, even though this higher level cutoff of HbA_1c further compromises its sensitivity. (25) The Australia AusDiab study recommended that HbA_1c < 5.5% be chosen to rule out diabetes and HbA_1c >= 7.0% be chosen to rule in diabetes as they found HbA_1c < 5.5% provided 99% negative predictive value and HbA_1c >= 7.0% provided 100% positive predictive value. (25) However, Raman et al reported only 24% PPV for HbA_1c >= 7.0%. (16) The lower PPV shows that there are genuine concerns about the variations in HbA_1c assay which need to be standardized. However, some of the variation could be due to the population and methodology being different.
A cross sectional population based Iranian study compared HbA_1c against FPG >=126mg/dl rather than full OGTT. They used the cutoff point of HbA_1c >= 6.0% recommended by the DCCT and found 86% sensitivity, 78% specificity, 36% PPV and 97% NPV. (18)

In India, Kumar et al reported an optimal cutoff similar to our study i.e. 6.1%, which gave 81% sensitivity and 81% specificity. (20) This lower cutoff increases the sensitivity of the test and avoids missing a

diagnosis of diabetes in a large number of cases. Further, use of a lower than optimal level to rule out diabetes and higher than optimal level to rule in diabetes can also be followed.

Although HbA_1c with a lower cutoff would have many advantages in screening a population without known diabetes, it has some disadvantages that limit its use. It is costly, has lower sensitivity, 2 hour PCPG is stronger than it in predicting CVD, standardization is often poor leading to unreliability of results, does not unveil within day disturbances in glucose metabolism and it is affected by haemolysis and ethnicity of the population. (26) As a consequence, in situations where performing the OGTT is not feasible, instead of using only FPG or only HbA_1c, a combination of both is more predictive than either parameter alone. (13)

The findings and implications of this study were limited by the fact that it was a hospital based study with a limited sample size and levels of HbA_1c were not correlated with clinical complications. Future community based study on newly diagnosed diabetics and obese prediabetics could test the sensitivity of a combination of fasting plasma glucose and HbA_1c against OGTT and correlate these with microvascular and macrovascular clinical complications.

V. Conclusion

In conclusion, the agreement between the two criteria- HbA_1c and mini-OGTT was good for diabetes but poor for pre-diabetes. HbA_1c proved to be slightly inferior but comparable to mini-OGTT in detecting diabetes. The use of the optimum cutoff of 6.1 found in this population is recommended to narrow this gap and improve the sensitivity of HbA_1c. HbA_1c also detected significantly more pre-diabetics than mini-OGTT, hence it may be used to screen the local undiagnosed population.

References

[1]. Kasheervar SA, Cornwall J. The current state of diabetes mellitus in India. Australas Med J. 2014; 7(1): p. 45-48.
[2]. Yang W, Lu J, Weng J, Jia W, Ji L, Xiao J, et al. Prevalence of diabetes among men and women in China. N Engl J Med. 2010; 362: p. 1090-101.
[3]. Ramachandran A, Mary S, Yamuna A, Murugesan N, Snehalatha C. High prevalence of diabetes and cardiovascular risk factors with urbanization in India. Diabetes Care. 2008; 31: p. 893-898.
[4]. Cockram CS, Lau JT, Chan AY, Woo J, Swaminathan R. Assessment of glucose tolerance test criteria for diagnosis of diabetes in Chinese subjects. Diabetes Care. 1992; 15(8): p. 988-90.
[5]. The International Expert Committee. International Expert Committee Report on. Diabetes Care. 2009; 32(7): p. 1327-1334.
[6]. McCance DR, Hanson RL, Pettitt D, Bennett PH, Hadden DR, Knowler WC. Diagnosing diabetes mellitus---do we need new criteria?. Diabetologia. 1997; 40: p. 247-55.
[7]. American Diabetes Association. Diagnosis and Classification of Diabetes. Diabetes Care. 2010; 33(1): p. S62-S69.
[8]. World Health Organization. Use of Glycated Haemoglobin (HbA1c) in the Diagnosis of Diabetes Mellitus:Abbreviated Report of a WHO Consultation. ; 2011.
[9]. Davidson MB, Schriger DL. Effect of age and race/ethnicity on HbA1c levels in people without known diabetes mellitus: implications for. Diabetes Res Clin Pract. 2010; 87(3): p. 415-21.
[10]. Ziemer DC, Kolm P, Weintraub WS, Vaccarino V, Rhee MK, Twombly JG, et al. Glucose-independent, black-white differences in hemoglobin A1c levels: a cross-sectional analysis of 2 studies. Ann Intern Med. 2010; 152(12): p. 770-7.
[11]. Tsugawa Y, Mukamal KJ, Davis RB, Taylor WC, Wee CC. Should the Hemoglobin A1c Diagnostic Cutoff Differ Between. Ann Intern Med 2012. 2012; 157(3): p. 153-159.
[12]. Herman WH, Yong MA, Uwaifo G, Haffner SKSE, Horton ES, Lachin JM. Differences in A1C by Race and Ethnicity Among Patients With. Diabetes Care. 2007; 30(10): p. 2453-2457.
[13]. Perry RP, Shankar RR, Finerberg N, McGill J, Baron AD. HbA1c Measurement Improves the Detection of Type 2 Diabetes in High-Risk Individuals With Nondiagnostic Levels of Fasting Plasma Glucose. Diabetes Care. 2001; 24(3): p. 465-71.
[14]. Hjellestad ID, Astor MC, Nilsen RM, Softeland E, Jonung T. HbA1c versus oral glucose tolerance test as a method to diagnose diabetes mellitus in vascular surgery patients. Cardiovasc Diabetol. 2013;: p. 12-79.
[15]. Incani M, Sentinelli F, Perra L, Pani MG, Porcu M, Lenzi A, et al. Glycated hemoglobin for the diagnosis of diabetes and prediabetes: Diagnostic impact on obese and lean subjects, and phenotypic characterization. J Diabetes Invest. 2015; 6: p. 44-50.
[16]. Raman PG, Maitra S. A comparative study of oral glucose tolerance test and glycated haemoglobin in high risk patients for diabetes mellitus. Int. J. Diab. Dev. Countries. 2000; 20: p. 23-27.
[17]. Riet ER, Alssema M, Rijkelijhhuizen JM, Kostense , P J, Nijpels G, et al. Relationship Between A1C and Glucose Levels in the General Dutch Population. Diabetes Care. 2010; 33(1): p. 61-66.
[18]. Ghazanfari Z, Haghdoost AA, Alizadeh SM, Atapour J, Zolala F. A Comparison of HbA1c and Fasting Blood Sugar Tests in General Population. Int J Prev Med. 2010; 1(3): p. 187-194.
[19]. Rivers KL, Hanna-Mahase C, Frankson MA, Peter S, Smith FP. Comparison between the Oral Glucose Tolerance Test and the Hba1c Assay for Detecting Impaired Glucose Regulation in Bahamian Adolescents. [Online].; 2015 [cited 2015 October 10. Available from: http://dx.doi.org/10.4172/2155-6156.1000511.
[20]. Kumar PR, Bhansali A, Ravikiran M, Bhansali S, Dutta P, Thakur JS, et al. Utility of Glycated Hemoglobin in Diagnosing Type 2 Diabetes Mellitus: A Community-Based Study. J Clin Endocrinol Metab. 2010; 95(6): p. 2832-35.
[21]. Engelgau MM, Thompson TJ, Herman WH, Boyle JB, Aubert RE, Kenny SJ, et al. Comparison of Fasting and 2-HourGlucose and HbA1c Levels for DiagnosingDiabetes. Diabetes Care. 1997; 20(5): p. 785-791.
[22]. Gomyo M, Sakane N, Kamae I, Sato S, Suzuki K, Tominaga M, et al. Effects of sex, age and BMI on screening tests for impaired glucose tolerance. Diabetes Res Clin Pract. 2004; 64(2): p. 129-36.
[23]. Anonymous. The diabetes control and complications trial and follow-up study, U.S. Department of Health and Human Services. ; 2008.
[24]. American Diabetic Association. Implications of the United Kingdom prospective Diabetes Study. Diabetes Care. 2000; 23(Suppl 1): p. S27-S31.
[25]. Florkowski C. HbA1c as a Diagnostic Test for Diabetes Mellitus –Reviewing the Evidence. Clin Biochem Rev. 2013; 34: p. 75-83.
[26]. Bonora E, Tuomilehto J. The Pros and Cons of DiagnosingDiabetes With A1C. Diabetes Care. 2011; 34(2): p. S184-S190.